

## AMENDMENTS TO THE CLAIMS

Please amend claims 1-3 and 8-10, add new claims 18 and 19, and cancel withdrawn claims 4, 5, 13, and 15, as shown in the following listing of claims, which will replace all prior versions and listings of claims in the application. Please cancel claims 4, 5, 13, and 14, which were previously withdrawn as non-elected claims, without prejudice to the filing of one or more divisional applications.

### Listing of claims:

1 (currently amended). A method for testing a test plasmid containing a gene encoding for an endothelial cell mitogen for the ability to produce a biologically active endothelial cell mitogen protein, wherein endothelial cells demonstrate enhanced survival in a cell survival assay in response to conditioned media from a transfection host cell line transfected with the test plasmid in comparison to conditioned media from a transfection host cell line transfected with a control plasmid, the method comprising:

- a. transiently transfecting a transfection host cell line with a test plasmid containing a gene encoding for an endothelial cell mitogen;
- b. incubating test sample endothelial cells with conditioned media from the transiently transfected transfection host cell line;
- c. determining the ability of the test sample endothelial cells to reduce MTS to formazan; and
- d. determining the level of cell survival of the test sample endothelial cells incubated with conditioned media from the transfection host cell line transfected with the test plasmid containing a gene encoding for an endothelial cell mitogen as compared to control sample endothelial cells incubated with conditioned media from the transfection host cell line transfected with a control plasmid;

wherein the level of cell survival of both the test sample and control sample endothelial cells is determined by the ability of the endothelial cells to reduce MTS to formazan;

and wherein the level of cell survival of the test sample endothelial cells indicates that the test plasmid produces a biologically active endothelial cell mitogen.

2 (currently amended). The method of claim 1, wherein the test plasmid contains a gene encoding for an endothelial cell mitogen selected from the group consisting of acidic and basic fibroblast growth factors, vascular endothelial growth factor, epidermal growth factor, transforming growth factor  $\alpha$  and  $\beta$ , platelet-derived endothelial growth factor, platelet-derived growth factor, tumor necrosis factor  $\alpha$ , hepatocyte growth factor and insulin-like growth factor.

3 (currently amended). The method of claim 2, wherein the test plasmid contains a gene encoding for VEGF.

4. – 5. (canceled)

6 (original). The method of claim 1, wherein the transfection host cell line is the Cos-1 cell line.

7 (original). The method of claim 1, wherein the endothelial cells are HUVEC cells.

8 (currently amended). The method of claim 1, wherein the level of cell survival of the test sample endothelial cells incubated with conditioned media from the transfection host cell line transfected with the test plasmid containing a gene encoding for an endothelial cell mitogen is at least 25 % fold greater than the level of cell survival of the control sample

endothelial cells incubated with conditioned media from the transfection host cell line transfected with the control plasmid.

9 (currently amended). The method of claim 1, wherein the test plasmid containing the gene encoding for the endothelial cell mitogen is tested for the ability to produce biologically active endothelial cell mitogen protein prior to use of the plasmid containing the gene encoding for the endothelial cell mitogen in a human gene therapy treatment.

10 (currently amended). A method for evaluating the ability of a first plasmid DNA construct containing a gene encoding for an endothelial cell mitogen to produce a bioactive endothelial cell mitogen protein as compared to the ability of a second plasmid DNA construct containing a gene encoding for an endothelial cell mitogen to produce a bioactive endothelial cell mitogen protein, wherein endothelial cells demonstrate enhanced survival in a cell survival assay in response to conditioned media from a transfection host cell line transfected with the first plasmid in comparison to a ~~transfection~~transfection-host cell line transfected with the second plasmid, the method comprising:

- a. transiently transfecting a first sample of a transfection host cell line with the first plasmid DNA construct containing a gene encoding for an endothelial cell mitogen;
- b. incubating endothelial cells with conditioned media from the transiently transfected transfection host cell line of step a;
- c. transiently transfecting a second sample of the transfection host cell line with the second plasmid DNA construct containing a gene encoding for an endothelial cell mitogen;
- d. incubating endothelial cells with conditioned media from the transiently transfected transfection host cell line of step c;
- e. determining the ability of the endothelial cells transfected with the first plasmid to reduce MTS to formazan in comparison with the ability of the endothelial cells transfected with the second plasmid to reduce MTS to formazan; and

f. determining the level of cell survival of the endothelial cells of step b incubated with conditioned media from the transfection host cell line transfected with the first plasmid containing a gene encoding for an endothelial cell mitogen as compared to the endothelial cells of step d incubated with conditioned media from the transfection host cell line transfected with the second plasmid containing a gene encoding for an endothelial cell mitogen;

wherein the level of cell survival of the endothelial cells is determined by the ability of the endothelial cells incubated with conditioned media from the first sample transfected with the first plasmid to reduce MTS to formazan in comparison with the ability of the endothelial cells incubated with conditioned media from the second sample transfected with the second plasmid to reduce MTS to formazan and

wherein the levels of cell survival of the endothelial cells indicate that the first plasmid produces an active endothelial cell mitogen.

11 (previously presented). The method of claim 10, wherein the first plasmid contains a gene encoding for an endothelial cell mitogen selected from the group consisting of acidic and basic fibroblast growth factors, vascular endothelial growth factor, epidermal growth factor, transforming growth factor  $\alpha$  and  $\beta$ , platelet-derived endothelial growth factor, platelet-derived growth factor, tumor necrosis factor  $\alpha$ , hepatocyte growth factor and insulin-like growth factor.

12 (previously presented). The method of claim 11, wherein the first plasmid contains a gene encoding for VEGF.

13. – 14. (canceled)

15 (original). The method of claim 10, wherein the transfection host cell line is the Cos-1 cell line.

16 (original). The method of claim 10, wherein the endothelial cells are HUVEC cells.

17 (original). The method of claim 10, wherein the plasmids containing the gene encoding for an endothelial cell mitogen are being compared as a means for determining an optimal plasmid construct for use in a human gene therapy treatment.

18 (new). A method of preparing a plasmid producing biologically active endothelial cell mitogen protein for use in a human gene therapy treatment, wherein the method comprises:

- a. preparing a test plasmid in a large quantity, wherein the test plasmid contains a gene encoding for an endothelial cell mitogen;
- b. testing the test plasmid according to the method of claim 1, wherein the test plasmid containing the gene encoding for the endothelial cell mitogen is tested for the ability to produce biologically active endothelial cell mitogen protein for use in a human gene therapy treatment.

19 (new). A method of preparing a plasmid producing biologically active endothelial cell mitogen protein for use in a human gene therapy treatment, wherein the method comprises:

- a. preparing a first plasmid in a large quantity, wherein the first plasmid contains a gene encoding for an endothelial cell mitogen;
- b. preparing a second plasmid in a large quantity, wherein the second plasmid contains a gene encoding for an endothelial cell mitogen;
- c. testing the first plasmid and the second plasmid according to the method of claim 10, wherein the first plasmid containing a gene encoding for the endothelial cell mitogen is tested for the ability to produce biologically active endothelial cell

mitogen protein and the second plasmid containing a gene encoding for the endothelial cell mitogen is tested for the ability to produce biologically active endothelial cell mitogen protein;

- d. comparing the results of the first plasmid and the second plasmid in step c as a means for determining an optimal plasmid construct for use in a human gene therapy treatment.